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University of the Philippines

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SAMPLE RICE PRODUCTION STATISTICS OF SELECTED TYPES OF CULTURE IN THE PHILIPPINES [1]

BASILIO N. DE LOS REYES

The long-felt need for an agency that will collect reliable agricultural statistics was recognized by our national leaders with the creation of the Division of Agricultural Economics (DAE) under the Department of Agriculture and Natural Resources in 1953. The Division is charged with the responsibility of conducting nation-wide surveys to gather data for preparing estimates of crop production especially rice. Scientific methods are employed in sampling and estimating actual yields and in forecasting production.

Other agencies like the Bureau of Agricultural Extension, National Economic Council, Bureau of Census and Statistics, etc., also collect information pertinent to rice production. These agencies supplement whatever information is available on rice production gathered by the DAE. These data are sometimes used for formulating policies in connection with the agencies' operations.

THE SURVEY

The Department of Agricultural Economics, U.P. College of Agriculture conducted a survey of 5,494 farms in the different areas of the Philippines in 1955. This survey covered results of farming operations for the crop year 1954-55. Although this was primarily intended to be a study of farm management, land use, and tenancy in the Philippines, essential information on rice production was also gathered.

The data were collected by staff members and trained students who were to write theses in agricultural economics. Each student was requested to interview 100 to 120 farmers in his respective barrio. If the number of farmers in the barrio was not sufficient, he was to interview additional farmers in the adjacent barrios. A sampling procedure was devised for each student. To have a better coverage

[1] General Contribution No. 886. Based on the paper read at the Symposium—Forum on the Statistics of Rice Production sponsored by the U.P. College of Agriculture on June 28, 1958, Manila.

of the country, staff members were sent to collect information in areas not covered by students. Staff members collected data from 90 farms in each province where they were assigned. Sample municipalities and barrios were selected in consultation with the provincial agriculturists.

The survey must not be considered as representing a cross-section of the region or province since the barrios were not randomly selected; thus, the data gathered are true reflections of only the barrios or towns. Neither can the data from this survey reliably represent rice production statistics for the province or region. The survey was designed for a number of objectives other than just to collect rice production statistics.

The surveys were mostly conducted in well developed areas which were accessible by roads or near the poblacion where students reside. Even the sample taken by the staff members may not reliably represent the provinces in which they were taken. The selection of the areas was decided on the basis of what the provincial agriculturists thought representative.

Furthermore, the sample was heavily weighted by farms from Central and Southern Luzon. About 68 per cent of the farms surveyed were in these two regions. The sample was also heavily weighted by the lowland rice farms which constituted about 72 per cent of the farms studied.

RICE PRODUCTION STATISTICS

Farms were divided into three groups based on the type of rice culture, as follows:

(a) Irrigated lowland rice farms—farms on which irrigation water was available throughout the year and two crops of rice were raised in a year under puddled conditions.
(b) Unirrigated lowland rice farms—farms dependent on rain water for irrigation and on which only one crop of rice was raised in a year under puddled conditions.
(c) Upland rice farms—farms in which rice was grown under the ordinary dry method. This includes the "kaingin" method.

Types of farms. The average yield of palay per hectare was highest on irrigated lowland rice farms (table 1). In these farms the average yield of palay per hectare was 44 cavans for the wet-season crop and 39 cavans for the dry-season crop. These farms

have a decided advantage over the other types because irrigation water is available throughout the year or at least during the growing period of the rice crop.

TABLE 1.—*Average yield of palay per hectare, selected provinces, Philippines, 1954-1955*

REGION AND PROVINCE	TYPE OF RICE CULTURE				ALL TYPES
	Irrigated Wet	Lowland Dry	Unirrigated Lowland	Upland	
Number of farms	814	814	2,820	1,403	5,037
Total area (ha.)	1,546	1,482	5,583	1,848	10,459
Ilocos	[a]	[a]	31	31	31
Abra	[a]	[a]	24	23	23
Ilocos Norte	—	—	31	[a]	31
Ilocos Sur	—	—	32	—	32
La Union	—	—	39	34	37
Cagayan Valley	[a]	[a]	32	—	32
Isabela	[a]	[a]	32	—	32
Central Luzon	41	33	42	30	40
Bataan	—	—	33	24	32
Bulacan	51	34	53	[a]	50
Nueva Ecija	—	—	53	36	52
Pampanga	37	31	34	[a]	33
Pangasinan	—	—	41	[a]	40
Tarlac	36	33	31	42	32
Zambales	—	—	26	[a]	25
Southern Luzon	54	48	39	21	38
Batangas	[a]	[a]	—	23	23
Cavite	—	—	34	20	32
Laguna	55	48	52	15	48
Quezon	[a]	[a]	26	—	26
Rizal	54	56	39	18	38
Bicol	33	32	32	18	31
Albay	31	32	21	16	30
Camarines Sur	50	36	33	25	35
Visayas	—	—	20	17	19
Capiz	—	—	25	20	23
Leyte	—	—	16	12	15
Mindanao	[a]	[a]	26	21	23
Agusan	—	—	26	27	27
Davao	[a]	[a]	26	[a]	24
Surigao	—	—	29	15	18
All Regions	44	39	38	23	36

[a] Less than 10 farms represented.

Unirrigated lowland rice farms ranked second in yield of palay per hectare. The average yield was 38 cavans per hectare.

The average production of 23 cavans per hectare of palay was lowest on upland palay farms. Upland palay is mostly grown where unfavorable topography or lack of developed water resources makes the flooding of rice fields impossible.

Regions and provinces. Among the areas studied, Central Luzon had the highest average production per hectare for all types of rice culture. The average yield per hectare was 40 cavans. Only Mindanao and the Visayas had average yields below 27 cavans per hectare. Mindanao had an average production of 23 cavans per hectare and the Visayas, 19 cavans per hectare.

On irrigated lowland rice farms, Southern Luzon had the highest average yield per hectare and the Bicol Region, the lowest. The average production in Southern Luzon was 54 cavans per hectare for the wet-season crop and 48 cavans per hectare for the dry-season crop. The average production in Southern Luzon was heavily weighted by farms from Laguna and Rizal.

On unirrigated lowland rice farms, Central Luzon had the highest average production per hectare, followed by Southern Luzon. The average production was 42 cavans per hectare for Southern Luzon. Bulacan and Nueva Ecija had the highest average production in Central Luzon and Laguna in Southern Luzon. The average production of 52 cavans per hectare in Laguna was only a cavan lower than that in Bulacan and Nueva Ecija.

In all the regions raising upland rice, only the Ilocos Region and Central Luzon had yields above 27 cavans per hectare. The average production for upland rice in the Ilocos Region was 31 cavans per hectare and for Central Luzon, 30 cavans per hectare.

CONCLUSIONS AND RECOMMENDATIONS

It is evident that there exists great variation among yields of palay by type of rice culture and by provinces and regions. Yields per hectare were highest on irrigated lowland rice farms and lowest on upland rice farms. Central and Southern Luzon had highest yields of palay per hectare than the other regions.

In collecting and reporting rice production statistics, there is a need for classifying farms by types of rice culture. The purpose is to make rice production statistics comparable with similar data in other countries. This will also provide a basis for formulating definite programs for increased rice production. It will show progress made and delineate areas for improvement.

BACTERIAL BLIGHT OF RICE[1]

AGUSTIN N. PORDESIMO[2]

WITH TWO TEXT FIGURES

The bacterial blight of rice in the Philippines was first reported by Reinking in 1918 as bacterial leaf stripe. Because the characteristic symptoms and signs of this rice disease are identical with the bacterial blight of rice reported in Japan by Ishiyama (1928), it is held probable that it is also caused by *Xanthomonas oryzae* (Uyeda and Ishiyama) Dowson.

The bacterium is claimed to differ from the forms of *Xanthomonas translucens* only by its failure to liquefy gelatin. The present study, however, shows that in parallel experiments with *Xanthomonas translucens, undulosa X. translucens* from rye grass and *X. citri*, it can liquefy gelatin. Since *X. oryzae* could not be distinguished from the members of the *translucens* group by morphological and physiological characters, the writer is proposing to subsume it under the *translucens* group and to give it the name *Xanthomonas translucens* (J.J.R.) Dowson f. sp. *oryzae* (Uyeda and Ishiyama) *n. comb.*

The disease is best transmitted artificially by first injuring the rice leaves with carburundum, then spraying on them a suspension of the inoculum. This procedure will be used in screening the susceptible varieties of rice.

Bacterial blight is a serious foliage disease of both upland and lowland rice (*Oryza sativa L.*) in the Philippines. It is usually more severe in upland varieties. The susceptible varieties may sustain considerable losses because extensive leaf area may turn yellow and dry up prematurely. Decreased functional foliage area may, in turn, cause stunting of the plants.

In September, 1956, a survey of the upland rice plots of the Plant Breeding Division, Department of Agronomy, U.P. College of Agriculture, for incidence of bacterial blight of rice showed that among the many susceptible varieties, Fortuna, a recommended upland variety, was most susceptible. Of the several resistant varieties,

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[1] Central Experiment Station Contribution No. 1982. Based on a portion of a thesis submitted to the Graduate Faculty of Cornell University in partial fulfillment of the requirements for the degree of Master of Science.

[2] The writer gratefully acknowledges his indebtedness to Dr. C. S. Reddy, former Cornell visiting professor of plant pathology, for advice in conducting the experiments and in the preparation of the manuscript; to Drs. G. O. Ocfemia and F. T. Orillo for suggestions in writing the manuscript; and to Messrs. L. N. Gibe and T. T. Reyes for assistance in the preparation of illustrations.

Texas Patna x B3812-A was completely free, Hill Medium Selection Maligaya, resistant, and Milfor, moderately resistant under field conditions.

The disease was first reported in this country by Reinking in 1918, but no detailed study on it has been made since that time. A similar disease of rice was reported in Japan by Uyeda and Ishiyama in 1922 and Ishiyama in 1928. They named the pathogen *Pseudomonas oryzae* Uyeda and Ishiyama; later, it was re-named by Dowson (1943) as *Xanthomonas oryzae* (Uyeda and Ishiyama) Dowson. It was held to be distinct from other form species of *Xanthomonas translucens* (J.J.R.) Dowson occurring in barley, speltz, rye, wheat, oats, brome grass, and timothy.

The present work aimed to (1) determine the relation of the pathogen causing bacterial blight of rice to a group of similar pathogens on cereals and other grasses in other countries, (2) ascertain whether or not the causal organism should belong to the *translucens* group of bacteria and if so, to amend the name, and (3) develop a technique for separating, by inoculation, the susceptible individual plants from the resistant ones for purposes of breeding for bacterial blight resistance.

OCCURRENCE AND SUSCEPT RANGE

The disease occurs principally on *Oryza sativa L.* throughout the Philippines. Bacterial stripe has been recorded by the Plant Breeding Division, U.P. College of Agriculture, as occurring at eight experiment stations in Luzon, Visayas, and Mindanao. No estimate of crop losses due to the disease has ever been recorded.

Goto, Fukazu, and Ohata (1953) reported that isolates from *Oryza sativa* L. were pathogenic to *Zizania aquatica* L., *Phalaris arundinacea* L., *Leersia oryzoides* (L.) var. *japonica, Phragmites communis* Trin., *Isachne globosa,* and other grasses and that isolates from *Z. aquatica* L. and *L. oryzoides* var. *japonica* were pathogenic to rice. Of these suscepts only *P. communis* Trin. (*P. vulgaris* [Lam.] Trin.), appears in *A Flora of Manila* (Merrill, 1912).

The writer's careful examination of grasses surrounding seriously infected rice plants and of grasses along the borders of rice fields, where bacterial blight was in epiphytotic form, indicated that the disease did not attack the naturally occurring grass species. The suscept range in the Philippines was not determined in the present study.

Aside from the Philippines and Japan, the bacterial blight of rice was also reported by Vzoroff as occurring on rice throughout the U.S.S.R. in Europe (Anonymous, 1938).

APPEARANCE OF THE DISEASE

Symptomatology. The first noticeable symptoms of the disease are water-soaked lesions in the leaves. The affected tissues soon lose their normal green color, passing through translucent yellowish to brownish tints. Translucency by transmitted light remains a consistent character of the lesions even in the later brown stage (fig.1).

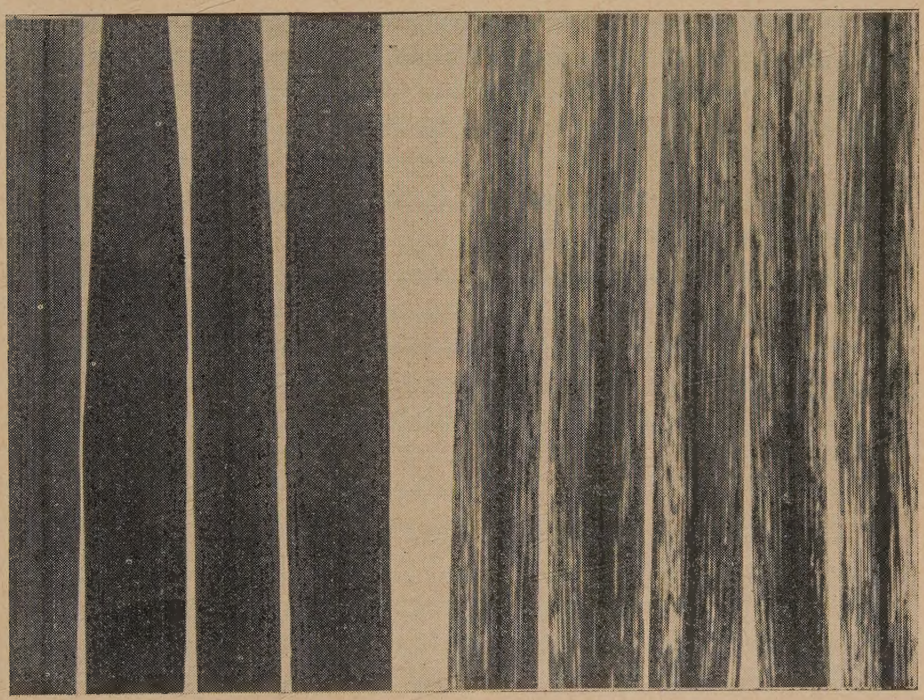

Fig. 1.—Bacterial blight of rice: natural infection showing the translucent character of the lesions in the later brown stage. Left, healthy leaves; right, naturally infected leaves. The photograph was made by using transmitted light to show the translucent character of the invaded areas. (Photograph taken by the Photographic Division).

The symptoms and signs of the disease conform to those described by Reinking (1918) and Ishiyama (1928); hence, it is believed that the disease in the Philippines is identical with the disease caused by *Xanthomonas oryzae* in Japan.

The characteristics, especially the translucent streaks, resemble closely those caused by all forms of *Xanthomonas tranlucens* occurring on barley, spelt, rye, wheat, oats, brome grass, and timothy (Jones, Johnson and Reddy, 1917; Reddy, Godkin and Johnson, 1924; Wallin and Reddy, 1945).

From published descriptions (Breed, Murray, and Hitchens, 1948; Elliott, 1951) it could be inferred that *Xanthomonas oryzae* (Uyeda and Ishiyama) Dowson differ only slightly and doubtfully from any of those in the *translucens* group.

The streaks vary from barely visible ones to those over two centimeters long on the leaf blades of the artificially inoculated seedlings. Blotching of the whole leaf was common on plants either artificially inoculated or naturally infected. These symptoms are produced on rice by inoculating with pure cultures isolated from bacterial blight-infected rice in the Philippines. In all instances, the progress of the water-soaked lesion is longitudinal; invasion is retarded or limited by the veins. Infections have not been observed on leafsheaths in the field or in the greenhouse.

Signs. Yellowish orange droplets of milky bacterial exudate appear on the surface of the lesion under humid conditions on warm days. Upon drying these droplets form hard resinous granules. This syndrome characterizes bacterial blight infection on cereals and grasses caused by all forms of *X. translucens.*

MATERIALS AND METHODS

A number of isolations were made from infected leaves showing the water-soaked appearance characteristic of bacterial blight lesions in the early stage of development. In all instances, a slow-growing waxy-yellow bacterium developed on the plates within three days at room temperature. Agar streaks were made, and plates poured from one of these agar streaks gave a large number of colonies of one type. Several pure cultures were obtained from these colonies. All proved to be identical organisms with the same cultural characters. Also, all proved to be pathogenic when sprayed on young rice plants. All control plants in these inoculation experiments remained healthy.

At various times the organism was also obtained from both fresh and dried exudates. Despite the admixture of other organisms,

the pathogenic species was readily separated from the others. Isolations were also made from exudates taken directly from diseased leaves in the field and also from plants artificially infected in the greenhouse.

The organism was also isolated from dried infected leaf kept for nine months. Small pieces cut from lesions were immersed directly in bouillon for ½ hour and dilution platings were then made from the bouillon. Pure cultures of the organism were obtained and their pathogenicity proven by inoculations.

Unless otherwise indicated, inoculation experiments were made on 3-leaf stage seedlings of the susceptible rice variety, Fortuna. A DeVilbiss No. 15 nasal type atomizer was used for spraying the plants with water dilutions of 72-hour-old bacteria grown on rice-decoction-agar slants. Several inoculation experiments will be described in the later part of this paper.

The studies on the cultural and physiological characters of the pathogen were based on 5 isolates, 3 of which were from naturally-infected leaves and 2 from artificially-infected plants of the variety Fortuna. The methods used were those described in the *Manual of Methods for Pure Culture Study of Bacteria* (Committee on Bacteriological Technic, 1923-1954). Studies on the cultural and physiological characters of the rice pathogen were made along with two pathogens in the *translucens* group, *Xanthomonas translucens undulosa* Hagborg 1942 (XT8) and *X. translucens* (?) from rye grass Z. Patrick 1951 (XT18),[3] and one other bacterial pathogen, *X. citri* for comparison in parallel cultures.

The pathogenicity of *X. translucens undulosa* was first tested on a Formosan wheat, variety Eho Chuchon [4] and *X. citri* on mandarin. The pathogenicity of *X. translucens* from rye grass was not tested due to lack of rye grass seeds.

All cultural and inoculation experiments were carried out under fluctuating tropical temperatures ranging from 26°C to 32°C in a refrigerator. However, gelatin-stab cultures were kept at constant temperature of 20°C in a refrigerator.

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[3] The cultures were supplied by Dr. W. H. Burkholder, Department of Plant Pathology, Cornell University, Ithaca, N.Y., U.S.A.

[4] Furnished by Mr. Ricardo M. Lantican, Division of Plant Breeding, Department of Agronomy, U.P. College of Agriculture.

THE PATHOGEN

Morphology

The description of *Xanthomonas oryzae* by Uyeda and Ishiyama (1922) shows that *X. oryzae* is almost identical in size and morphology with the first form of *X. translucens* isolated and described by Jones, Johnson and Reddy (1917).

Cultural characters

Agar poured plates. On beef-extract-peptone agar at 28°C, colonies appeared in 60 to 72 hours and at the end of 4 days they were 1 to 2 mm. in diameter, circular, smooth and shining. The margin was entire. Colonies were wax-yellow in color, soft but not viscid in consistency. Buried colonies were small and disc-shaped.

Gelatin plates. In 7 days at constant temperature of 20°C inside the refrigerator, surface colonies showed a characteristic margin which may have been due to liquefaction of the medium. In 20 days liquefaction was complete and colonies floated.

Agar slants. On beef-extract-peptone-agar slants, streak cultures in 48 hours made a moderate, filiform, wax-yellow, opaque, smooth, shining, convex growth. Early growth was butyrous but later it became hard and difficult to remove with a needle. All isolates grew slowly on this medium.

Rice-decoction agar. In 48 hours, streak-culture growth was moderate, filiform, convex, glistening, dull yellow, butyrous, and the medium was unchanged. Streak cultures kept inside the refrigerator exhibited similar characteristic growth in 72 hours.

Potato-dextrose agar. Growth was copious, filiform, wax yellow, glistening, convex, butyrous; medium was unchanged.

Nutrient broth. In beef-extract-peptone bouillon, uniform clouding occurred within 48 hours. This clouding subsequently became rather strong; sediment was flocculent. Ring was formed on surface growth.

Physiological characters

Gelatin stabs. At 20°C in nutrient gelatin and in beef-extract-peptone gelatin, the surface growth was abundant after several days with a medium pit of liquefaction. At the end of 10 days, at constant temperature of 20°C in the refrigerator, the growth became napiform and slowly liquefied its way toward the bottom of the tube.

The degree of liquefaction of gelatin by *Xanthomonas oryzae* was about the same as *X. translucens undulosa* during this period. Both progressed somewhat less than halfway to the bottom of the tube in 20 days as shown in figure 2 which represents the typical form of liquefaction on the basis of average measurements of 10 stab cultures.

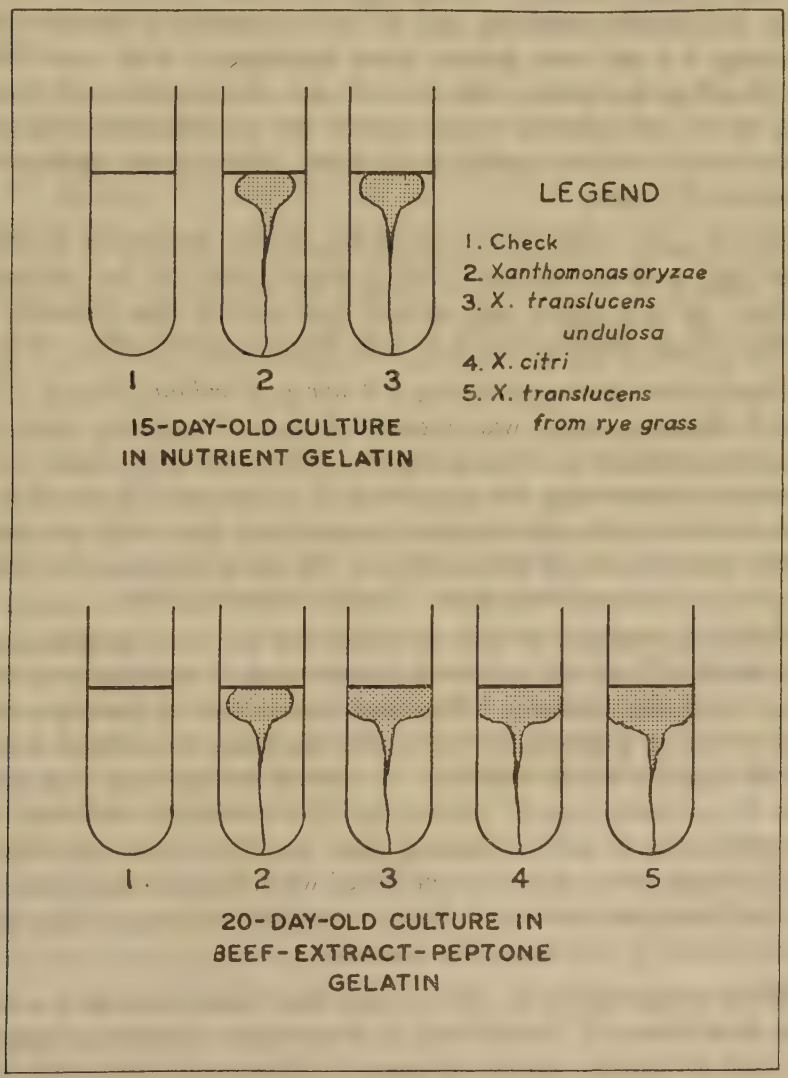

Fig. 2.—Typical forms of gelatin liquefaction by four species of *Xanthomonas* based on average measurements of 10 stab cultures for each species. (Photograph taken by the Photographic Division).

In parallel experiments, inoculated tubes of gelatin kept under ordinary room temperature (26°C to 30°C) were no longer capable of solidifying when they were transferred to the refrigerator after 20 days. Uninoculated controls congealed.

Gelatin-plate streaks. Three-day-old streak cultures of *X. oryzae, X. translucens undulosa,* and *X. citri* on plates of nutrient agar containing 0.4 per cent gelatin were surrounded with clear zones after flooding the plates with 8 to 10 ml. of a solution of 15 gm. $HgCl_2$ in 100 ml. distilled water and 20 ml. concentrated HCl. The checks formed a white opaque precipitate. Clear zones indicated liquefaction of gelatin.

Starch agar. Starch hydrolysis was hardly detectable by transmitted light without the aid of white paper placed at the bottom of the dish. A very slight halo around the colonies was detected immediately after flooding the dish with dilute Lugol's iodine solution.

Beef-extract agar containing 0.2 per cent soluble starch. Evidence of diastatic action was observed 5 days after using dilute Lugol's iodine solution as test for starch. Clear zones of about 3 to 4 mm. radius surrounding the colonies of *X. oryzae* and *X. translucens undulosa* were barely detectable by transmitted light with the aid of a white paper placed at the bottom of the dish, whereas, the dishes of *X. citri* were completely clear. Checks remained blue.

Partial hydrolysis of both to which 0.2 per cent soluble starch had been added was also observed after a week of incubation at fluctuating room temperatures. This was determined by placing a drop of the culture in a depression on a porcelain plate and adding a drop of dilute Lugol's iodine solution. A reddish-brown color was noted in the drops containing *X. oryzae* and *X. translucens undulosa;* the drop containing *X. citri* remained clear and the check was stained blue. This reaction was very pronounced when the determination was done on 2-week-old cultures. There is meager literature describing the reaction of *X. oryzae* on this medium.

When compared to *X. citri,* which the writer considers a very strong hydrolyzer, *X. bryzae* and *X. translucens undulosa* showed a very weak hydrolytic action on starch. This corroborates the findings of Reddy, Godkin, and Johnson (1924) that the *translucens* group of bacterial blight organisms "produced a very feeble diastatic action on starch noticeable for approximately one-half hour after

iodine solution had been applied. After that time it was difficult to detect any halo about the colonies."

INOCULATION EXPERIMENTS

The disease was reproduced with the characteristic symptoms by artificial inoculations of rice and the organism recovered by re-isolation.

The first inoculation was performed on September 27, 1956. Rice seeds of Fortuna variety were sown in 6-inch pots. When the seedlings were in the 3-leaf stage, 10 uniform pots, each containing about 15 plants, were selected for experimental use. Of these, 2 pots designated as (I) were inoculated by spraying the uninjured leaves with a suspension of the organism prepared by macerating newly infected rice leaves and allowing the bacteria to diffuse into the water. The suspension was rid of bits of leaves by pouring it through two thicknesses of clean cheese-cloth into the DeVilbiss atomizer. Another set of 2 pots (II) were inoculated by spraying the plants with bacterial suspension prepared by pouring sterile water into 3-day-old agar tube streak cultures and the bacterial growth allowed to diffuse into the water. Another 2 pots (III) were first dusted with 320-mesh carborundum before spraying the bacterial suspension prepared as in II. The plants in another 2 pots (IV) were dusted with carborundum as in III, followed quickly by drawing the leaves gently between the thumb and index finger, and sprayed with the bacterial suspension prepared as in II. The plants in pot V were dusted with carborundum, the leaves gently drawn between the thumb and index finger and sprayed with sterile water. The plants in pot VI were uninjured and sprayed with sterile water. Pots V and VI served as checks.

The 10 pots of rice plants were placed outdoors before and after inoculation; the weather was warm (27°C) and moderately humid (78 per cent relative humidity). Early in the morning and late in the afternoon the leaves were covered with dew.

Except in the checks (pots V and VI) infection was noted nine days after inoculation in all the inoculated pots. Ten days after inoculation, pot IV showed approximately 95 per cent of the plants infected, III about 50 per cent, II approximately 30 per cent, and I about 15 per cent. Plants in pots V and VI remained free from bacterial infection.

In another series of experiments conducted on October 23 and November 2, 1956, severe infection occurred on Fortuna seedlings that were inoculated as in IV above, whereas, the check plants remained healthy. Earliest noticeable symptoms in these experiments were observed 4 days after inoculation.

Other abrasives. On February 20, 1957, two other abrasives were compared with carborundum in inoculation work. These were white earth and diatomaceous earth that were previously ground and passed through U.S. Standard Sieve No. 60. These materials were separately dusted by means of a DeVilbiss 118 duster. Fifteen pots, each containing 25 one-month-old Fortuna seedlings grown in the open were used. Three pots were dusted with carborundum; another 3 pots with white earth; and the other 3 pots with diatomaceous earth. Leaves of all dusted plants were drawn between the thumb and index finger and with an atomizer inoculated with the bacterial suspension from 3-day-old streak cultures on rice-decoction agar. For the control, 6 pots were left undusted and uninjured. Of these 6 pots 3 were sprayed with the inoculum and the other 3 pots with sterile water. Inoculation was done late in the afternoon, the temperature ranging from 26°C to 28°C. All plants were kept outdoors.

Seven days later on February 27, severe infections were observed on all carborundum-dusted seedlings. There was no infection on either the white- or diatomaceous-earth dusted plants and none on the check plants. There was slight infection on the inoculated undusted plants.

On March 1 and 5, 1957, the same abrasives were used but this time an electric sprayer (Hudson Lektrik-Spray, Wizard Sprayer) instead of a DeVilbiss atomizer was used to spray the inoculum prepared as previously mentioned. Inoculation was done outdoors late in the afternoon, the temperature ranging from 29°C to 31°C. Thirteen-day-old Fortuna seedlings were used. Experimental and check plants were kept in the open before and after inoculation since greenhouse temperature was higher than outdoor temperature. Again, carborundum-dusted plants showed 100 per cent infection 9 days after inoculation. All other inoculated plants showed below 50 per cent infection. Check plants remained healthy.

Also, on the above dates, these abrasives were added separately to the bacterial suspension at the rate of 2 grams per 100 ml. suspension. An electric sprayer was used in spraying the suspension of abrasives and inoculum; however, the results were no better than

those obtained with the carborundum as used in the previous experiments.

Electric sprayer vs. DeVilbiss atomizer. The electric sprayer and the DeVilbiss atomizer were used to spray inoculum on plants dusted with the 3 abrasives mentioned and also to spray the abrasive-inoculum combination on plants not dusted. The results indicated that the electric sprayer is not superior to the DeVilbiss atomizer in inoculating 3-leaf-stage potted seedlings.

In general, slight infection was obtained from the foregoing inoculation experiments. This may have been due to the high temperature which caused rapid drying of the spray droplets. Results of these experiments, however, showed that infection was easily secured under normal environmental conditions when inoculations were made in conjunction with the use of abrasives to artificially rupture the epidermis of the suscept. Carborundum consistently increased the percentage of infected plants in all artificial inoculations.

Dipping trimmed seedlings. On March 23, 1957, 30-day-old potted Fortuna seedlings were trimmed and dipped in a heavy suspension of bacterial inoculum containing a detergent, Shellestol (Shell Co.), used at the rate of 1.5 ml. per liter of suspension. This material was proven by motility test to be non-toxic to this bacterial pathogen. Infection was not noted even after the emergence of the next two succeeding leaves. Plants that were trimmed but whose trimmed edges were dipped in sterile water likewise remained uninfected. Seedlings of the same age that were dusted with carborundum and the leaves gently drawn between the thumb and index finger before spraying them with the inoculum-detergent combination prepared previously mentioned were abundantly infected when observed nine days later. Check plants that were dusted and rubbed with carborundum and sprayed with sterile water plus detergent remained uninfected.

Inoculation in relation to stomatal movement. The inoculum was prepared from 3-day-old agar tube cultures shortly before inoculation at hourly intervals starting from 8:30 a.m. to 4:30 p.m. The inoculated and check plants were exposed to direct sunlight before being sprayed; these were returned to the greenhouse benches after exposure. The highest percentage of infection was obtained on plants that were sprayed with the inoculum in suspension at 11:30 a.m. on March 23 and on April 6, 1957. At that time of the

day, majority of the stomata were presumably open. Check plants remained healthy.

Seed inoculation. On March 23, 1957, Fortuna seeds previously soaked for 48 hours in sterile water and rinsed several times with sterile water were coated with a heavy suspension of the inoculum and allowed to germinate in vermiculite. Soaked but uncoated seeds were used as checks. Germinated seedlings both in the checks and the inoculum-coated seeds remained free from infection even after 2 weeks.

In another set of experiments, Fortuna seeds were held in running water overnight and then rinsed three times with sterile water. These seeds were planted directly on 3-day-old bacterial colonies in Petri dishes. Seeds planted on uninoculated plates served as checks. All seeds on the checks as well as on the inoculated plates germinated without any bacterial infection. The data were insufficient to permit definite conclusions on whether or not this method could be useful in routine inoculation tests.

Of all the inoculation methods used in the experiments, inoculation of carborumdum-injured seedlings with the aid of an atomizer appears to be the most effective for producing infections. This method being the easiest to perform will be tested for screening susceptible varieties and lines of rice. Only those with a fairly high degree of resistance are likely to remain uninfected. Plants subjected to this severe test that escape infection are almost certain to possess a high degree of resistance under field conditions. Preliminary results with this method indicate that it will be effective in eliminating the more susceptible individuals.

TAXONOMIC CONSIDERATIONS

In published accounts, the bacterial pathogen occurring on rice in Japan was originally named in 1922 as *Pseudomonas oryzae* Uyeda & Ishiyama (Uyeda and Ichiyama, 1922). It was later named *Bacterium oryzae* (Uyeda and Ichiyama) Nakata in 1928. In 1937 it was named *Phytomonas oryzae* (Uyeda & Ishiyama) Magrou and in 1943, Dowson adopted the name *Xanthomonas oryzae* (Uyeda & Ishiyama) Dowson (Elliot, 1951).

Probable synonyms as found in Bergey's Manual are *Pseudomonas itoana* Tochinai, 1932; *Bacterium itoanum* Burgwitz, 1935;

and *Phytomonas itoana* Magrou, 1937 (Breed, Murray, and Hitchens, 1948).

Published reports describe *Xanthomonas oryzae* (Uyeda & Ishiyama) Dowson differently from other forms of *X. translucens* mainly in one physiological character, i.e., liquefaction of gelatin which is reported negative for the rice organism. Starch hydrolysis was not reported by Ishiyama (1928) but undoubtedly, this is one of the two physiological characteristics that was used to separate *X. oryzae* from the *translucens* group of bacterial blight organism on cereals and other grasses in temperate countries.

In these studies, it was found that *X. oryzae* positively exhibited hydrolysis of starch although to a very slight degree. Parallel experiments with *X. translucens undulosa* and *X. translucens* on rye grass showed a similar reaction. This characteristic of the forms of *X. translucens* which had been reported negative at one time (Jones, Johnson and Reddy, 1917) was later found to be positive (Reddy, Godkin and Johnson, 1924).

Although liquefaction of gelatin by *X. oryzae* appeared in the literature as negative, it proved to be positive as shown by several trials in parallel experiments with *X. translucens undulosa* and *X. translucens* on rye grass. The tests showed that *X. oryzae* liquefies gelatin in much the same way as do the two forms of *X. translucens* already mentioned. Consequently this character cannot be used to separate *X. oryzae* from the *translucens* group of bacterial blight organisms.

Previous investigators found that the pathogens of the *translucens* group differ in no way from each other morphologically, culturally, and physiologically. They separated members of the group on the basis of pathogenicity by the use of trinomials. It is now shown that *X. oryzae* is also in this group.

For clarification and for the convenience of plant pathologists, a very important phenomenon to consider in placing *X. oryzae* in a new position is the host which is attacked. Because there exists an earlier-published valid epithet for the group to which *X. oryzae* is identical in morphology and physiology, and since the author of *X. oryzae* did not adopt this earliest legitimate epithet available for the group with its particular circumscription, position and rank, the writer is of the opinion that the oldest legitimate name of the rice pathogen, *X. oryzae*, should be in combination with the oldest legitimate epithet, *X. translucens*.

These studies prove that *X. oryzae* belongs in the *translucens* group. Therefore, to conform with the naming of members of this group such as, *Xanthomonas translucens* f. sp. *undulosa* (Smith, Jones, Reddy) Hagborg, it is proposed to lower the species name to a form species so that *X. oryzae* becomes *Xanthomonas translucens* ([J.J.R.] Dowson) f. sp. *oryzae* (Uyeda & Ishiyama) n. comb.

LITERATURE CITED

ANONYMOUS. 1938. Department of Plant Pathology. Rep. Del. Agr. Exp. Sta. 1937–1938 (Bul. 214), pp. 29-45, 3 figs. (*Abst. from* R.A.M. *18:* 379).

BREED, R. S., E. G. D. MURRAY, and A. P. HITCHENS. 1948. Bergey's manual of determinative bacteriology. 6th ed. xvi + 1529 pp. Baltimore, Maryland: Williams and Wilkins Co.

Committee on Bacterial Technic. 1923-1954. Manual of methods for pure culture study of bacteria. Geneva, N.Y.: Biotech Publ.

DOWSON, W. J. 1943. On the generic names *Pseudomonas*, *Xanthomonas*, and *Bacterium* for certain bacterial plant pathogens. Trans. Brit. Mycol. Soc. **26:** 4-14.

ELLIOTT, CHARLOTTE. 1951. Manual of bacterial plant pathogens. 2nd ed. viii + 186 pp. Waltham, Mass: Chronica Botanica Co.

GOTO, K., R. FUKAZU, and K. OHATA. 1953. Overwintering of the causal bacteria of rice leaf blight in the rice plant and grasses. Agri. and Hort. **28:** (1): 207-208. (*Abs. from* R.A.M. **32:** 506).

ISHIYAMA, *S.* 1928. Bacterial leaf blight of the rice plant. Proc. Third Pan-Pacific Sci. Congr., Tokyo, 1926, p. 2112.

JONES, L. R., A. G. JOHNSON, and C. S. REDDY. 1917. Bacterial blight of barley. J. Agr. Res. (U.S.) **11:** 625-644.

MERRILL, E. D. 1912. A flora of Manila. 489 pp. Manila, Philippines. Mimeographed.

REDDY, C. S., J. GODKIN, and A. G. JOHNSON. 1924. Bacterial blight of rye. J. Agr. Res. (U.S.) **28:** 1039-1040.

REINKING, OTTO. 1918. *Oryza* sativa Linn. Rice: Bacterial leaf stripe. *In* Philippine economic-plant diseases. Philippine J. Sci. **13:** 225-226.

WALLIN, J. R., and C. S. REDDY. 1945. A bacterial disease of *Phleum pratense* L. Phytopathology **35:** 937-939.

UYEDA and S. ISHIYAMA. 1922. *Pseudomonas oryzae;* Mitteil. Kaiserl. Centralb. Landw. Versuchsst. Tokyo **45:** 233-261.

BIOCHEMICAL STUDIES ON ABACÁ MOSAIC AND BUNCHY-TOP VIRUSES: III. ORGANIC ACIDS AND MOSAIC VIRUS INFECTION [1]

APOLINARIO R. CARANGAL, JR., ERLINDA A. TUCAY and FERNANDO A. BERNARDO

The leaves of the plants susceptible to the abacá mosaic disease had generally lower amounts of oxalic acid than the leaves of the resistant plants. The non-inhibitory action of oxalic acid on mosaic infection and the fact that mosaic infection does not affect the level of organic acids (acetic, oxalic, lactic group and formic group) in abacá leaves suggest that differences in the amount of oxalic acid of susceptible and resistant plants are not at all related to the disease.

An earlier study of the organic acids of abacá by Carangal *et al.*[2] reported that oxalic, acetic and lactic or succinic acids are present in the roots, stems and leaves of normal and diseased abacá plants. Apparently abacá mosaic virus infection does not cause abnormal metabolism of these acids through the formation of acids other than those found in healthy plants. The work was continued to include other plants used in the pathological and breeding studies of the abacá mosaic disease, with emphasis on the oxalic acid content and the quantitative aspects of organic acid metabolism.

EXPERIMENTAL PROCEDURE

Determination of organic acids. Leaves from susceptible and resistant plants were analyzed for their oxalic acid content. Susceptible plants included *Musa textilis,* abacá; *M. textilis x M. banksii,* "Alinsanay"; *M. banksii,* "Saguing maching"; *Zea mays,* corn; *Canna indica;* arrowroot; *M. textilis x M. balbisiana,* "Minay"; *Ensete glaucum;* ornamental canna.

Resistant plants selected were canton, pacol, abacá × pacol, *Musa balbisiana,* "Saba", and *M. accuminata,* "Agotay".

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[1] Central Experiment Station Contribution No. 1983. Research Project No. 800. Prepared in the Department of Agricultural Chemistry and Division of Plant Breeding.

[2] A. R. Carangal, Jr., A. M. Catambay and E. B. Manzanilla, *Philippine Agriculturist,* **39**, 17 (1955).

The leaves were dried in a forced-draft oven at 80°C for 4 to 6 hours, ground in a Wiley mill and stored in a desiccator preparatory to analysis.

The dried sample from each plant was analyzed for oxalic acid content by weighing and boiling with 50 ml. 1 N H_2SO_4 for 2 hours. Water was added occasionally. The boiled samples were filtered and the filtrate together with the washings was made up to volume. Aliquots were then concentrated to a 1 to 2 ml.-volume which was used in the subsequent chromatographic separations of the oxalic acid.

The acid was eluted from a silica gel column according to the procedure of Bulen *et al.*[3] The eluate corresponding to the point of emergence of oxalic acid was titrated with 0.01 N NaOH. Results were expressed in number of milliequivalents per gram of dried samples. Two runs were made for each sample.

Chemotherapy with oxalic acid. Suckers from diseased plants were selected and divided into two lots. Half of the lot was placed in 0.1 N oxalic acid solution. To facilitate the absorption of oxalic acid, portions of the suckers were cut under the solution. After being immersed for 3 days in the solution, the suckers were planted beside the untreated suckers. Treated and untreated plants were then planted in soil and given identical growing conditions. They were observed for incidence of mosaic symptoms and analyzed for organic acids.

Another attempt to cure the plant of the mosaic disease was made by using juice of sour "balimbing" (*Averhoa carambola*) as a form of oxalic acid solution. The ground fruits (equal to one-fourth the weight of the sand) were incorporated in the sand and mosaicked abacá seedlings were planted in the mixture. The plants were similarly observed for the characteristic mosaic symptoms. The organic acid content of these plants were analyzed after one month.

DISCUSSION OF RESULTS

The predominantly susceptible species like abacá, "alinsanay", "saguing maching," corn, canna and arrowroot had relatively low contents of oxalic acid, ranging from 5.74 to 33.35 milliequivalents per 100 grams of dry weight (table 1). The resistant species (canton, pacol, abacá × pacol hybrid, saba and agotay) had a higher

[3] W. A. Bulen, J. E. Varner, and R. C. Burrell, *Analyt. Chem.*, **24**, 187 (1952).

range—36.97 to 61.35. This is one evidence pointing to a possible role of oxalic acid in preventing inroads of the mosaic virus.

The complementary test, in which diseased suckers were infiltrated or steeped in either oxalic acid solution or the expressed juice of "balimbing", did not give any indication that the diseased condition was alleviated or eliminated. This may be taken to mean that oxalic acid cannot inhibit the multiplication of the mosaic virus inside the host. It cannot, however, be taken as conclusive because it was not ascertained whether or not the treatment effected the introduction of oxalic acid into the cells of the abacá suckers.

TABLE 1.—*Oxalic acid content of the leaves of plants susceptible and resistant to the abacá mosaic disease*

PLANT	NUMBER OF PLANTS	OXALIC ACID IN MEQ. × 10^2 PER GM. DRY WEIGHT		
		Min.	Max.	Ave.
Susceptible:				
Abacá, *Musa textilis*	10	17.56	41.26	30.47
"Alinsanay," *M. textilis* × *M. banksii*	6	27.67	36.27	31.42
"Saguing maching," *M. banksii*	8	20.44	49.22	33.35
Corn	4	3.74	8.00	5.74
Canna indica	6	15.78	18.87	16.82
Arrowroot	7	10.46	35.54	21.33
Ensete glaucum [a]	8	43.32	75.48	55.40
"Minay," *M. textilis* × *M. balbisiana* [a]	4	24.12	33.06	31.09
Ornamental canna [a]	6	38.58	56.08	49.79
Resistant:				
Canton	6	26.82	48.38	36.97
Pacol	6	45.50	67.90	56.93
Abacá × Pacol	4	35.76	48.69	42.25
"Saba," *Musa balbisiana*	8	41.72	56.67	50.84
"Agotay," *M. accuminata*	5	50.47	65.24	61.35

[a] These plants are less susceptible than abacá, the symptoms appearing after repeated inoculations.

Preliminary analysis of a limited number of diseased and healthy plants susceptible to the disease gave variable results as shown in table 2. The diseased seedlings of "Minay" (*M. textilis x M. balbisiana*) had a lower level of oxalic acid in their leaves than the healthy plants. On the other hand, the leaves of diseased canna, arrowroot and "Alinsanay" (*M. textilis x M. banksii*) had higher amounts of oxalic acid content than the leaves of the healthy seedlings.

TABLE 2.—*Effect of mosaic virus infection on the oxalic acid content of the leaves of susceptible plants*

PLANT	OXALIC ACID IN MEQ. × 10^2 PER GM. DRY WEIGHT [a]	
	Healthy	Diseased
Abacá	30.26	28.42
Corn	5.74	2.89
"Alinsanay," *M. textilis* × *M. banksii*	29.75	36.61
"Minay," *M. textilis* × *M. balbisiana*	31.09	21.12
Canna indica	17.82	25.98
Arrowroot	11.46	15.06

[a] Average of the analysis of four different seedlings.

In abacá seedlings grown under the same environmental conditions the level of the different acids, acetic, formic group, lactic group, and oxalic acid was not affected by mosaic virus infection. The leaves of both normal and diseased seedlings had comparable amount of these acids.

TABLE 3.—*Effect of mosaic virus infection on the organic acids of the leaves of abacá*

SAMPLE	MILLIEQUIVALENTS OF ACIDS × 10^2 PER GRAM DRY WEIGHT			
	Acetic	Formic	Lactic	Oxalic
Healthy				
1	13.4	2.5	2.2	34.4
2	10.8	1.0	0.9	31.1
3	7.0	1.0	0.8	35.4
4	8.4	1.6	0.7	22.3
5	12.2	1.8	0.4	45.6
6	13.6	1.2	0.2	34.2
Average	11.07 ± 2.17	1.51 ± 0.45	.87 ± .45	33.83 ± 4.47
Diseased				
1	14.0	4.1	0.5	35.0
2	10.1	1.1	0.5	35.0
3	20.3	2.0	0.9	30.2
4	10.7	1.5	0.3	26.8
5	17.8	1.8	0.2	29.6
6	12.9	1.0	0.5	45.3
Average	14.29 ± 3.16	1.89 ± .75	.47 ± .16	33.0 ± 4.83

Although plants susceptible to abacá mosaic disease had lower levels of oxalic acid in their leaves than the resistant plants, the failure to establish any relation between mosaic infection and this acid and the inconsistent levels of oxalic acid in healthy and diseased plants weigh heavily against its possible value as a diagnostic tool for determining resistance or susceptibility. Inhibition of the infection by oxalic acid was not established and its level in the leaves was not affected by virus infection. Such results may be related to the relatively inert nature of oxalic acid in organic acid metabolism and interconversion (Burris, 1953).[4] Oxalic acid is probably just an end product of the organic acid metabolism in the abacá plants.

[4] R. H. Burris, *Ann. Rev. of Plt. Physiol.*, 4, 91 (1953).

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Like a good architect, a researcher should have his whole plan down on paper. If the plan were carefully drawn, the second stage, implementing it, should not present any major problem at all. True, there are many circumstances and exigencies which cannot be anticipated (e.g., losing subjects in the course of the research). These for the most part are not irreparable or of too disastrous proportions. There will be more of such major unforseen difficulties for the beginning researcher than for the experienced one. It is, in fact, a hall-mark of good planning to be able to anticipate as many of these differences as is humanly possible.

—Rene V. Dawis

THE EFFECT OF SUPPLEMENTING ANTIOXIDANT IONOL-CP (2-6-DITERTIARY-BUTYL-P-CRESOL) TO A BROODING RATION FOR CHICKS [1]

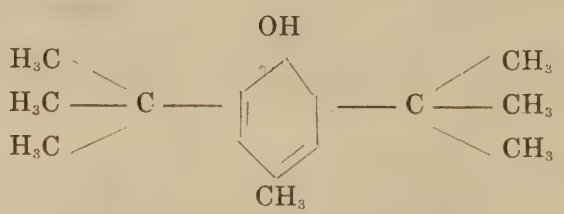
G. O. Ranit, R. Z. Ortigas and M. M. Ylagan

WITH TWO TEXT FIGURES

Significant improvement in growth was obtained for S. C. White Leghorn chicks fed a practical ration supplemented with 25 milligrams of Ionol-CP (2,6-di-tertiary-butyl-p-cresol) as an antioxidant. Increasing the level of the antioxidant caused a gradual decrease in growth rate but a gradual increase in the vitamin A content of the liver. Regression coefficients showed that an increase of 1 mg. of Ionol-CP per 100 grams of feed decreased the cockerel and pullet weights by 0.519 and 0.497 grams respectively but increased the vitamin A stored in the liver of males by 0.271 International Units per gram of liver.

The use of antioxidants in poultry feeding is widely practised in the United States. Matterson (1955) reported that the antioxidants, DPPD (N, N-diphenyl-p-phenylene diamine), BHT (2, 6-di-tertiary-butyl-P-cresol), and Santoquin (6-ethoxy-1, 2-dihydro-2, 2, 4-trimethyl-quinoline) prevented encephalomalacia, a disease in chicks caused by vitamin E deficiency. He also reported that the utilization of vitamin A in the body of chicks is improved by preventing the destruction of unsaturated fats and fatty acids through oxidation. Lease *et al.* (1938) demonstrated that oxidation of fats destroyed vitamin A in the guts of rats.

Ionol-CP (2, 6-di-tertiary-butyl-p-cresol, known also as 4-methyl-2, 6-ditertiarybutylphenol, and as ditertiarybutylhydroxytoluene) is an antioxidant with the following formula:



[1] Central Experiment Station Contribution No. 1984. Research Project No. 1192. Financed partly by a grant from the Shell Company of the Philippines, Ltd. Ionol-Cp was supplied through the courtesy of Mr. F. A. Maclang of said company.

It is used for preserving fats, oils, waxes, resins, and polyethelene films, and also for stabilizing dehydrated alfalfa, and vitamin preparations. Deichmann *et al.* (1955) observed no signs of intoxication or gross histopathological changes in dogs fed a daily dose of 0.17 to 0.94 gm. of ditertiarybutyl methylphenol (DBMP) for every kilogram of feed over a period of 12 months. Rats fed rations containing 0.5 or 1.0 per cent of this antioxidant in their diet over a period of 20 to 90 days did not find the feed palatable. In poultry, a level as low as .0125 per cent ditertiarybutyl paracresol (BHT) in the diet prevented experimentally-induced encephalomalacia (Bunnel *et al.*, 1955), increased the storage of carotene and vitamin A in the blood serum and liver of chicks (Potter *et al.*, 1956), and in the body and eggs of hens without adversely affecting their health, production and reproduction up to as high a level as 0.125 per cent of the diet (Shellenberger *et al.*, 1957). Other results of these studies led the writers to conclude that the protective action of antioxidants takes effect not only in the feed during storage but principally in the body of the chicks after the feed has been consumed.

The present study was conducted in the Poultry Husbandry laboratory of the U.P. College of Agriculture to determine the effect of supplementing a practical brooding ration with graded levels of Ionol-CP on the growth, livability and vitamin A storage in the liver of White Leghorn chicks.

EXPERIMENTAL PROCEDURE

Varying concentrations of Ionol (0, 25, 50, 100, 200, and 400 milligrams per 100 grams of feed) were added to a brooding ration consisting of the following ingredients with all parts by weight:

Rice bran	25.0
Ground yellow corn	44.7
Soybean oil meal	15.0
Fish meal	10.0
Ipil-ipil leaf meal	3.0
Aurofac	0.5
Ground oyster shell	1.0
Cod-liver oil	0.3

The calculated composition of the feed was as follows (per cent):

Protein	21.3
Fat	6.4

Fiber	4.3
Calcium	1.2
Phosphorus	0.7

The feeds were not stored before feeding. Each concentration was fed continuously for 7 weeks to 30 straight-run, day-old White Leghorn chicks hatched from the College flock. The chicks were housed in electrically heated battery brooders for the first four weeks, then transferred to rearing cages that had wire floods and sun porches. The chicks in all the lots were given identical care and management. Weekly weights and feed consumption data were recorded.

At the end of the experiment, two cockerels in each lot were analyzed for vitamin A in the liver following the procedures outlined by The Association of Vitamin Chemists Incorporated (1951). Data obtained in this study were analyzed statistically using standard procedures (Snedecor, 1946).

RESULTS AND DISCUSSION

A summary of the data on the response of chicks fed rations supplemented with different levels of Ionol is shown in table 1.

TABLE 1.—*Effect of graded levels of Ionol fed on the growth, feed consumption, feed conversion, mortality and the Vitamin A content of the liver*

MG. OF IONOL/ 100 GM. FEED	AVERAGE BODY WEIGHT IN GM.		AVERAGE FEED CONSUMPTION IN KG.	FEED CONVERSION	PER CENT MORTALITY	I. U. OF VITAMIN A/GM. LIVER
	Male	*Female*				
0	353.3	320.6	1.026	3.36	23.3	43.5
25	445.0 [a]	389.7 [a]	1.055	2.79	13.3	65.5 [a]
50	397.0	367.3	1.089	3.15	30.0	67.7 [a]
100	356.4	314.8	1.093	3.50	30.0	77.1 [b]
200	329.7 [b]	309.3	0.926	3.20	13.3	132.8 [b]
400	228.8 [b]	189.3 [b]	0.763	4.65	6.7	152.3 [b]

[a] Significant.
[b] Highly significant.

Weight. Analysis of variance of the individual cockerel and pullet weights showed that the birds fed the 25 milligrams of Ionol per 100 grams of feed were significantly heavier than the controls, while those fed the ration containing 400 milligrams of Ionol per 100 grams were lighter than the controls. Further analysis revealed a highly significant regression of the 7-week body weight on the le-

vels of the antioxidant in the feed (fig. 1). Regression coefficients of –0.519 and –0.497 for cockerels and pullets respectively were obtained. An increase of 1 milligram of Ionol per 100 grams feed decreased the cockerel weight by 0.519 gram, and the pullet weight by 0.497 gram. The regression analysis was carried out only among the birds receiving antioxidant in the feed, since the growth response of the chicks between the 25- to 400-milligram level was entirely different from that between the zero and 25-milligram level. These

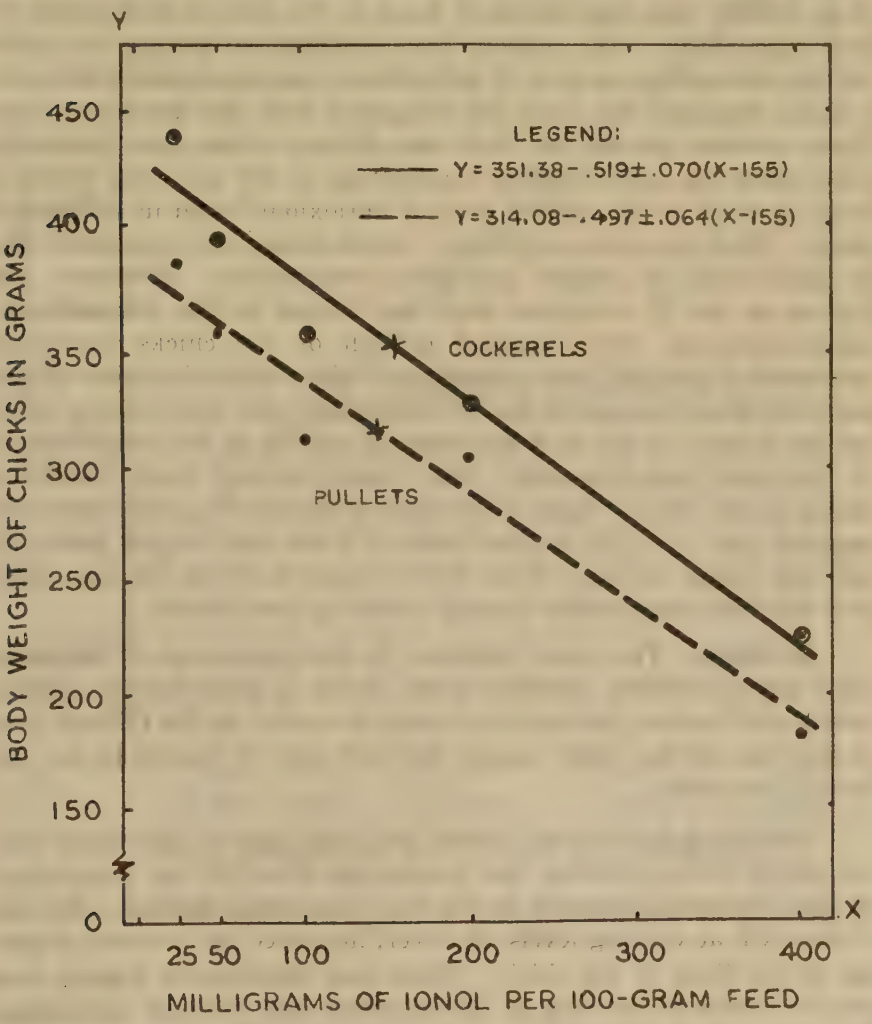


Fig. 1.—Regression of body weight of 7-week-old chicks on the level of Ionol in the feed.

results showed that the addition of the antioxidant at the 25-milligram level promoted growth of chicks. Increasing the antioxidant in the ration up to 400 milligrams to every 100 grams of feed retarded gradually the growth of the chicks.

Feed consumption. The level of Ionol in the feed affected the rate of consumption. The birds which received 200 and 400 milligrams of Ionol to every 100 grams of feed consumed less than those fed the lower levels. This result confirmed the claim of Deichmann *et al.* (1955) that high levels of Ionol in the diet of rats makes the feed unpalatable. The reduced feed consumption among the chicks fed the 400-milligram level of antioxidant supplementation indicated a lower tolerance for Ionol-CP compared with the laboratory rats. These workers pointed out that rats did not refuse feed containing Ionol until the concentration reached up to 0.8 per cent which is twice as much as the highest level of antioxidant used in the present study. Feed conversion efficiency varied among the treatments due to differences in weight and feed consumption. Efficiency was highest at the 25-milligram level and lowest at the 400-milligram supplementation. The retarded growth of the chicks cannot be attributed to reduced feed consumption alone since the rates of consumption from the zero to the 100-milligram level were almost equal; yet the weight of the bird decreased gradually as the concentration of the Ionol was increased. This relationship of body weight of chicks on the level of Ionol in the feed is presented in the regression analysis (fig. 1). The higher levels of Ionol used in this study did not only retard the growth of White Leghorn chicks but also rendered the feed unpalatable thereby reducing feed intake.

Mortality. The great variation in the percentage of mortality could not be entirely ascribed to the levels of Ionol tested. Cannibalism and feather picking which were prevalent in the College flock at the time of the study caused the high rate of deaths in the different treatments.

Vitamin A in the liver. Since the feeds used in this study were not stored before feeding, the preserving effect of the antioxidant for the oxidizable nutrients in the feed was rather limited. The use of Ionol-CP at varying levels, however, improved the vitamin A storage in the liver of the chick. There was significantly higher vitamin A content in the liver of the cockerels that received antioxidant than those that did not receive antioxidant. The increase in the amount of Ionol resulted in a corresponding rise in vitamin A in

the liver. A regression coefficient of +0.271 was calculated from the data, i.e., an increase of 1 mg. of Ionol per 100 gm. of feed was accompanied by an increase of 0.271 International Units of vitamin A stored in every gram of liver (fig. 2).

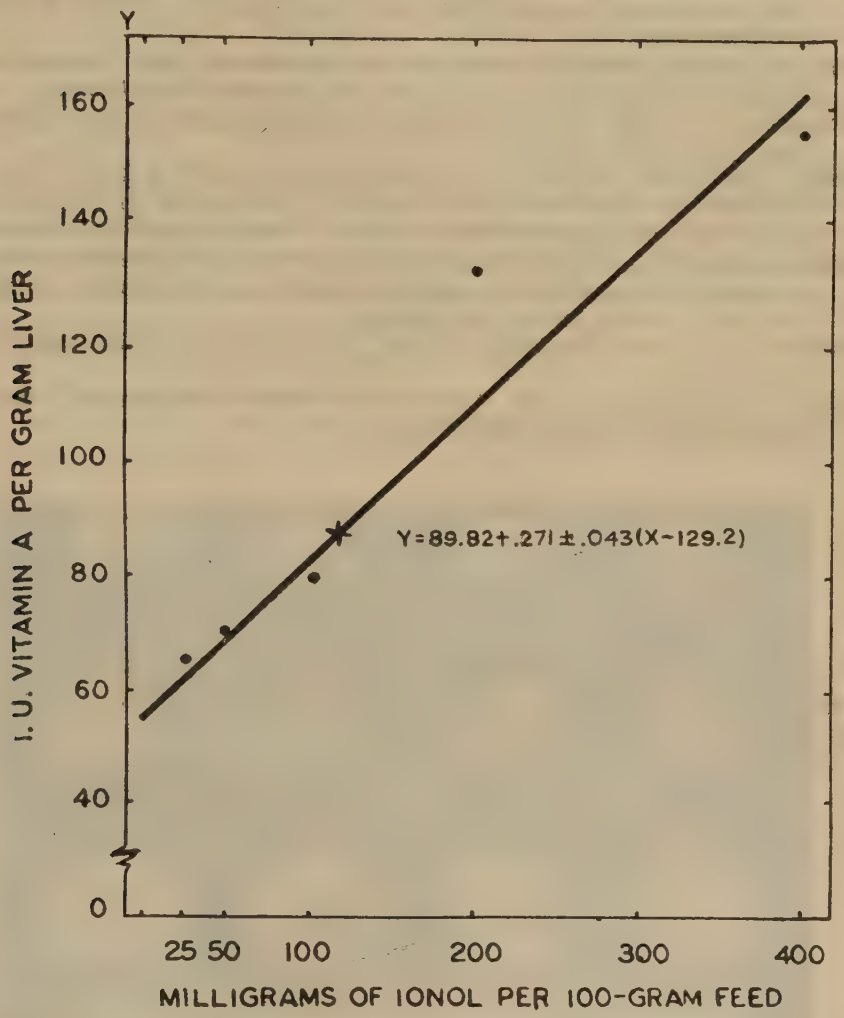


Fig. 2.—Regression of vitamin A content in the liver on the level of Ionol in the feed.

LITERATURE CITED

BUNNELL, R. H., L. D. MATTERSON, E. P. SINGSEN, L. M. POTTER and ANNA KOZEFF and E. L. JUNGHERR. 1955. Studies on encephalomalacia in the chick. 3. The influence of feeding or injecting various tocopherols and other antioxidants on the incidence of encephalomalacia. Poultry Science **34**: 1068-1075.

LEASE, E. J., J. G. LEASE, J. WEBER, and H. STEENBOCK. 1939. Destruction of vitamin A by rancid fats. Journal of Nutrition **16**: 571-583.

MATTERSON, L. D. 1955. Antioxidants—their present and potential use in feeds. Poultry Comment **12**: No. 1.

POTTER, L. M., R. H. BUNNELL, L. D. MATTERSON and E. P. SINGSEN. 1956. The effect of antioxidants and a vitamin B_{12} concentrate on the utilization of carotenoid pigments by the chicks. Poultry Science **35**: 452-456.

SHELLENBERGER, T. E., D. B. PARISH and P. E. SANFORD. 1957. Effects of antioxidants, DPPD and BHT, on health, production and reproduction of laying hens. Poultry Science **36**: 1313-1321.

SNEDECOR, G. W. 1946. Statistical methods. Ames, Iowa: The Iowa State College Press.

Methods of Vitamin Assay. The Association of Vitamin Chemists Inc. 1951. New York: Inter-science Publishers Inc.

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Science is not the whole of life: it does, however, offer careers of great satisfaction giving opportunity for creative research yielding, perhaps, results of world importance.

—*Sir John Cockroft*

NOTES ON MUTANT FORMS AMONG PHILIPPINE PLANTS I. A CASE OF GREEN SANTOL (*SANDORICUM KOETJAPE* MERR.)

D. L. UMALI and F. A. BERNARDO

WITH ONE TEXT FIGURE

A santol tree in Bilog-bilog, Tanauan, Batangas belonging to Mrs. Gregorio Perea was reported to be bearing fruits which remain green when ripe. The abnormal case was observed for two fruiting seasons, and the following data were collected:

Plant characters—Tree, about 10 years old and approximately 11 meters in height; trunk, about a foot in diameter; young branches and leaves, softly pubescent; flowers numerous, about a centimeter long and greenish yellow; fruits globose, about 4 centimeters in diameter when the tree is heavily bearing and 6.8 centimeters when the tree is poorly bearing; courge green when ripe.

Months of flowering—February to March

Eating quality—Very acidic

The plant does not differ from ordinary santol trees except in the color of ripe fruits (fig. 1).

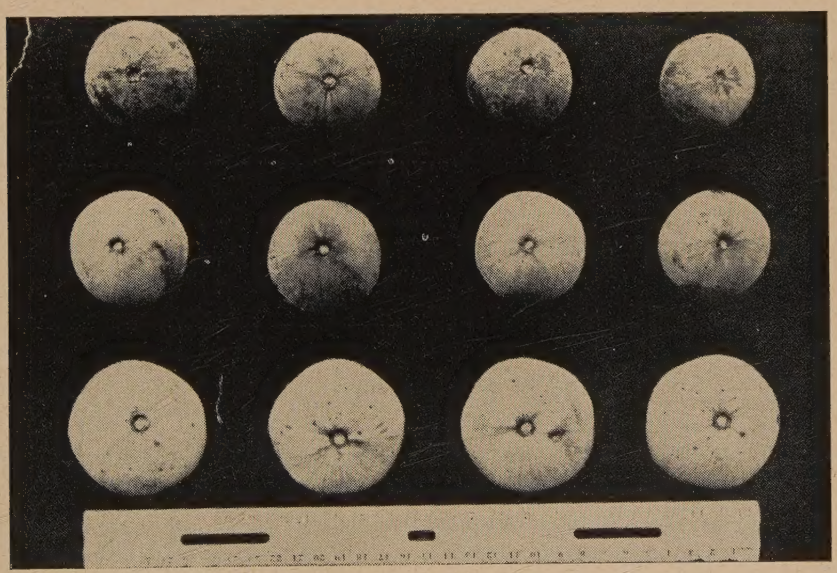

Fig. 1.—Ripe santol fruits: Upper row—Courge green [a] (uncommon); middle row—Ochraceous buff [a] (common); lower row—Warm buff [a] (common).

[a] Based on R. Ridgway, "Color standard and color nomenclature," A. Hoen & Co., Baltimore, Md., pp. 43, (1912).

ESTIMATING LIVEWEIGHTS FROM BODY MEASUREMENTS IN BERKJALA

J. A. Eusebio

Determination of liveweight is an important consideration in selecting breeding stock and in marketing swine. Since most swine raisers do not possess scales, they often resort to guesswork in estimating liveweights. Estimates may, however, be obtained from body measurements like volume of the body, height at withers, heartgirth and body length. Some of these measurements were reported to be highly correlated with weight (Eusebio *et al.*, 1957)[1]. As a result of the study these writers evolved the following formulae:

(1) $W = 207 - 4.7922X + 0.33963X^2$

where W stands for the estimated liveweight in kilograms and X for the heartgirth in centimeters. Heartgirth is the circumference of the body at the chest immediately behind the point of the elbow.

(2) $W = 8.5 - 0.670629L + 0.0117395L^2$

where L stands for the body length in centimeters. Body length is measured along the arch of the back from the poll, or the point midway between the ears, to the base of the tail. In making the measurement the head of the animal should be held in the normal position as shown in figure 1.

To illustrate the use of the formulae, we may take a heartgirth of 80 centimeters as an example. By substituting 80 for X in equation (1) and getting the algebraic sum of the terms, we arrive at 41.0 kilograms as the estimated liveweight. On the other hand, if 80 is the body length the estimated weight from equation (2) is 29.9 kilograms. In some cases estimated weights less than 30 and greater than 150 kilograms were found to be appreciably different from the actual weights. In other words, the equations are not reliable below 30 kilograms and beyond 150 kilograms. This is not deemed a serious limitation because farmers rarely keep butcher hogs long after they exceed the 100-kilogram weight, and pigs lighter than 90 kilos are not yet ready for the market.

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[1] J. A. Eusebio, G. B. Garcia and S. C. Salmon, *Philippine Agriculturist*, **41**, 252-260 (1957).

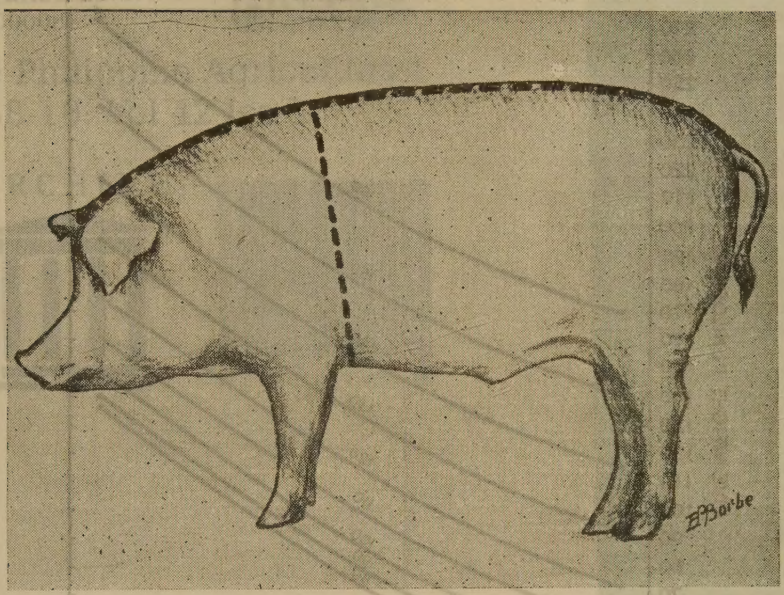

Fig. 1.—The head should be held in its normal position. The body length is measured along the arch of the back from the poll or the point midway between the ears to the base of the tail. The heartgirth is the circumference of the body at the chest immediately behind the point of the elbow.

The use of the equations require some familiarity with algebraic operations and many farmers are not trained in algebra nor are they inclined to perform algebraic operations. In order to provide an easy means of estimating liveweights, values for various pairs of heartgirths and body lengths were derived and the data were set in tabular form (table 1) and in the form of a graph (fig. 2).

Let us assume that an animal has a heartgirth of 100 and a body length of 120. By following the line of 120 in table 1 until it intersects with the column of 100, we arrive at 82 kilograms as the estimated weight. Measurements may be rounded off to their closest ten (for example 85 may be rounded off to 80 and 86 to 90) and arrive at a fairly rough estimate; a closer liveweight estimate may be obtained by interpolation. Using the heartgirth of 100 in the foregoing example, let us assume that the body length is 125. This is 5/10 of the interval between 120 and 130. The difference of the estimated weight (94 − 82) is 12. The estimated liveweight will be 82 + (5/10 × 12) = 88 kilograms.

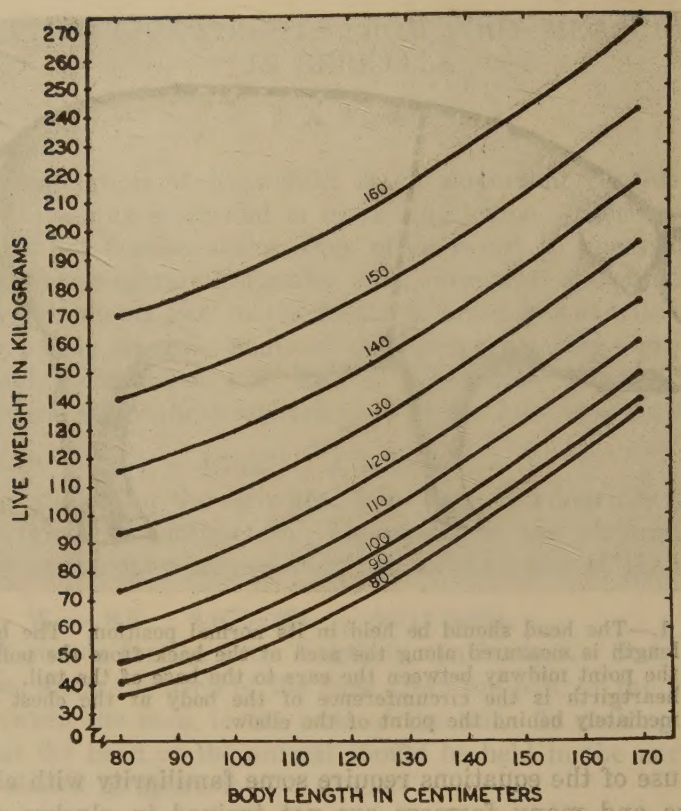

Fig. 2.—Graphical system of calculating liveweight.

In estimating the weight by the graph, we project a vertical line at point 120 on the base line until it cuts the curve corresponding to 100. Then the point of intersection is projected horizontally until it cuts the vertical coordinate. This will be at 82 kilograms, the estimated liveweight.